THE LARVA OF SARCOPHAGA, A PARASITE OF CISTUDO CAROLINA AND THE HISTOLOGY OF ITS RESPIRATORY APPARATUS.

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The student of zoölogy is early impressed with the intensive manner in which animal life has penetrated every available space. Even so remote and strange place as the poison glands of the rattle-snake have been entered, these glands furnishing ample proteid and oxygen supply for a little nematode that makes them its habitat. In the example of this *Sarcophaga* we find a fly that has entered the nucha of the "box-turtle"—a region of the body where its larva will not be exposed to serious pressure between parts of the "turtle's" body and where it will also be free from the attacks of the appendages and mandibles of the host.

The occurrence of this parasite in *Cistudo* was first observed by Packard ('82). Packard described and figured it as in œstrid larva. Thus, so far as I have been able to determine, arose the basis for believing that a "bot-fly" infested a reptile. Aldrich ('05) in referring to Packard's paper states that perhaps it is not an œstrid. Sharpe in the Cambridge Natural History says that Œstrididæ may occur in the reptiles. Wheeler ('90) next records the occurrence of the dipteron larvæ on the nucha of *Cistudo Carolina*. He succeeded in getting the larvæ to pupate and in rearing imagines from the pupæ. These adult flies proved to belong to the genus *Sarcophaga* and not to be œstrid flies. Thus there appears to remain no evidence of a "bot-fly" infesting a reptile.

In October, 1910, a female specimen of *Cistudo Carolina* was brought into my laboratory. It was kept through the winter in a sink. January, 1911, a student called my attention to what he called a "growth" in the nucha of the right side. This, however, proved to be an insect larva. Two days later the larva escaped from the perforation made in the skin of the host.

Subsequently two other larvæ left the excavated region of the nucha. These specimens were preserved in alcohol. May 14, the fourth and most vigorous larva dropped from the host. This larva was placed upon soil in a box where it burrowed into the earth and formed an oval, dark brown pupa. This pupa has not yielded an imago, so that I have been unable to corroborate Wheeler's diagnosis as based upon the adult fly.

Except for some details which are readily overlooked in preserved specimens, such as Packard had, the larvæ I found closely resemble the figures and descriptions given by Packard. With the living material which I had at my service, I was able to see details which make these larvæ correspond more closely to the following description of larvæ of Sarcophagidæ than to that of Œstrididæ larvæ. Brauer ('83) says that the larvæ of Sarcophagidæ "are rounded, thinner anteriorly and amphipneustic. The antennæ are short, thick, cylindrical, divergent, wart-like tubercles, each with two ocellus-like chitinous rings at the tip. The mouth hooklets are distinct, strongly curved and separated from each other. The abdominal segments are distinctly differentiated by transverse swellings and are each provided with a girdle of spines. The hind stigma-plate is situated in a deep cavity, which is formed by the last segment alone. The anal swelling is two-pointed. The puparium is oval.[1]" Thus I am led to infer that I have the same kind of larva that Packard had figured and described and am able to corroborate Wheeler's statement that this is not a "bot-fly" larva but a sarcophagid larva.

Apart from this I have been interested in certain details that no one has recorded for this particular sarcophagid. Figure 1 represents the dorsal aspect of the larva magnified ten diameters. Each segment is seen to bear a band of spines. The antennæ are seen from the ventral side (Fig. 6, *ant.*) together with the strongly curved, distinct mandibles (Fig. 6). On the ventral side of the posterior segment there is a trilobed disc armed with stout spines (Fig. 3 and Fig. 5, *d*). This may function as a sucking disc. The posterior end of the last segment is divided

[1] This translation of Brauer's description was taken from Williston's "North American Diptera," 3d ed., page 349, by Dr. J. M. Aldrich.

into a wide, dorsal lobe and a narrow, projecting, ventral lobe. Between these two lobes is a deep recess into which the anus and posterior stigmata open. The posterior stigmata are guarded by a large stigma-plate which has two lobes. Each lobe bears three spatulate chitinous bars (Fig. 4, *c.p.*) which articulate with six similar bars on the ventral lobe of the segment (Fig. 4, *c'.p'.*). The shape and relation of these dorsal and ventral chitinous bars to each other are such that I am led to believe that they function as prehensile structures; the lower lobe of the segment pressing its bars against the bars of the stigmatic plate can lay hold of the wall of the excavated region in the skin of the host and thus anchor the larva. The most striking feature to which attention has not been called is the presence of two anterior stigmata (Fig. 1, *st.*). These stigmata are fan-shaped structures which bear seventeen or eighteen papillæ along their terminal edge (Fig. 2, *st.*). In a specimen cleared with xylol each of these stigmata can be seen to lead directly into a large lateral trachea. Thus they are provided with an air-breathing apparatus though they live in a thick fluid of suppurated matter which makes liable the clogging of one or more of these tracheal openings or may necessitate the temporary closing of one of them. In this connection it is interesting to find a transverse tracheal commissure posterior to the anterior stigmata and another transverse tracheal commissure anterior to the posterior stigmata. These commissures enable both tracheal trunks to get air though for any reason some of the stigmata may be closed. Thus the chief tracheal system consists of a pair of anterior and a pair of posterior stigmata and two lateral tracheal trunks which are connected by means of an anterior and a posterior tracheal commissure.

Nothing unusual has been noted concerning the histology of the tracheal trunks and posterior stigmata. The histology of the anterior stigmata has, however, attracted my attention. These fan-shaped structures are for the most part proliferated masses of cuticle. The anterior half of the stigma projects beyond the contour of the body as a stigmatic process. The posterior half lies beneath the surface of the body and is covered by an epithelium which represents the hypodermis modified as tracheal epithelium (Fig. 8, *te.*). From the posterior margin of

the stigmatic process there is a cuticular and hypodermal invagination which extends to near the base of the stigma as a retaining thread (Fig. 8, *inv.*). This retaining thread of cuticle and hypodermal epithelium is seen in transverse section at *inv.* in Fig. 9. The entire stigma represents a modified region of hypodermis and cuticle. On the mesial side of the stigma near the base of its anterior third the hypodermis becomes very pronounced, the cells becoming very large and columnar. These cells, so far as their form is concerned, are the most conspicuous tracheal cells (Fig. 7, *te.*). From them slender processes go into the cuticular mass of the stigmatic process. These processes and the position of these cells suggest that they not only help to elaborate the cuticular substances of the stigmatic process but that, also, they may be able to move the stigmatic process. Within the mesial wall of the stigmatic process no cytoplasm extends except that of these cellular processes; within the lateral wall of the stigmatic process scattered hypodermal cells are found. There is thus an indifferent cellular supply to the tracheal process of the stigma. Indeed the entire stigma is for the most part a cuticular structure. The cuticle of the general surface of the body is distinctly two-layered. The outer layer is the deeper and in hæmotoxylin stains the more deeply. The inner layer is clearly a softer substance and does not stain deeply. These two strata are involved in the formation of the anterior stigma. The inner layer, except for becoming more abundant in the stigma, is not modified. Figure 7 at *c* and Fig. 8 show this layer of the cuticle passing over into that of the stigma. The outer layer of cuticle, however, is thinner over the stigmatic process than over the general surface of the body. When it reaches the tips of the papillæ it is invaginated and passes as a series of converging tubules to the bases of the papillæ where the tubules unite to form a large tube whose lining is confluent with the lining of the tracheal trunk. The cuticular lining of the tracheal trunk also presents a deeply staining layer and a layer that does not readily stain (Fig. 11, *tl.*), thus resembling the cuticle, of which I believe it represents a modified region. The inner denser layer of this tracheal lining gives rise to spiral tænidia as shown in Fig. 11 at *t.* When this denser layer passes

into that of the stigma very minute slender processes arise from it into the lumen: these processes branch and rebranch to form a reticulated layer which takes the place of the tænidia of the trachea (Fig. 10, *r.*). This reticulated layer is increased until the entire lumen is filled with a reticulated mass or plug (Figs. 8, 7, and 9, *rp.*). At the base of each papilla the reticulated plug branches and continues to near the tip of the papilla where there is a small chamber into which the branch of the reticulated plug sends its terminal filaments (text-figure 1). Thus we find

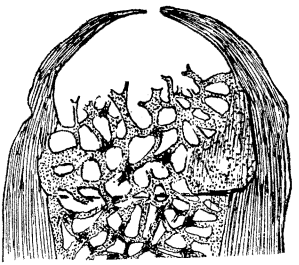

FIG. 1. Longitudinal section of a papilla of the anterior stigma, showing the terminal chamber into which filaments of the reticulated plug project. × 1,500.

the cuticle, tracheal lining and the cuticular mass of the stigma to be two-layered. In all three places the non-staining layer is little modified; but in the tracheal lining the deeply staining layer is modified to form the tænidia, and in the tracheal process it becomes a reticulated plug.

The larvæ of blow-fly and house fly have likewise prothoracic stigmatic processes with finger-like papillæ. These in turn, according to de Meijere ('02), have reticulated plugs which he calls "felt-chambers" (Feltkammern). What does such histological structure mean? We see the cuticular hairs guarding the stigmata of ants or other insects and we interpret them as being devices to protect the trachea from foreign bodies. But here we have in place of protecting hairs an extensive, finely reticulated plug which resembles the cotton plug of a bacterial culture tube as though it were constructed for the purpose of protecting the trachea from microscopically minute bodies. The larva feeds upon the suppurated fluid found within the excavated region of the nucha of the host, hence while the larva is feeding these bacteria can hardly be of service, for the anterior end of its body

is bathed in the suppurated mass. However, when about to pupate the larva reverses its position with reference to the suppurated mass, and lies with its anterior end directed towards or through the opening in the skin of the turtle. The larva is then in a position to breathe air through the anterior stigmata. At the same time the larva during the three or four days spent in emerging from the host, frequently retreats into the excavated cavity when disturbed, thus its anterior end may repeatedly become contaminated with the bacteria of the suppurated mass. I think, therefore, that the anterior stigmata are chiefly functional during the two or three days spent by the larva in passing from the turtle to the ground and that the reticulated plug is a bacterial screen protecting the trachea from infection threatened by the repeated retreat of the larva into the excavated cavity when it lies with its posterior end at or within the suppurated mass. If this conjecture concerning the time and character of the functioning of the anterior stigmata is not warranted, I believe that I am justified in agreeing with Hewitt ('08), that the anterior stigmata of this character are functional at some stage in the life of the larva.

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Explanation of Plate I.

Fig. 1. Dorsal aspect of larva. *st.*, stigma. × 10.

Fig. 2. Lateral aspect of anterior end of larva. *mo.*, mouth; *m.*, mandible; *st.*, stigma. × 100.

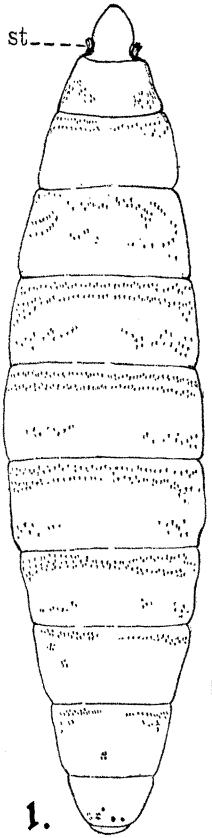
Fig. 3. Ventral aspect of posterior segment. *d.*, tri-lobed disc with stout spines. × 25.

Fig. 4. Ventral aspect of posterior segment. The ventral lobe is laid back so as to expose its six chitinous bars *c'p'.*, and the two-lobed stigma-plate with its six chitinous bars *cp.* × 25.

Fig. 5. Lateral spect of posterior end of larva. *d.*, tri-lobed disc; *cp.*, chitonous bar of stigma-plate. × 10.

Fig. 6. Ventral aspect of anterior end of larva. *m.*, mandible; *mo.*, mouth; *st.*, stigma; *ant.*, antenna. × 25.

Fig. 7. Transverse section through base of tracheal process at level indicated by arrow 7 on Fig. 8. *c.*, cuticle; *rp.*, reticulated plug; *h.*, hypodermis; *te.*, tracheal epithelium. × 250.



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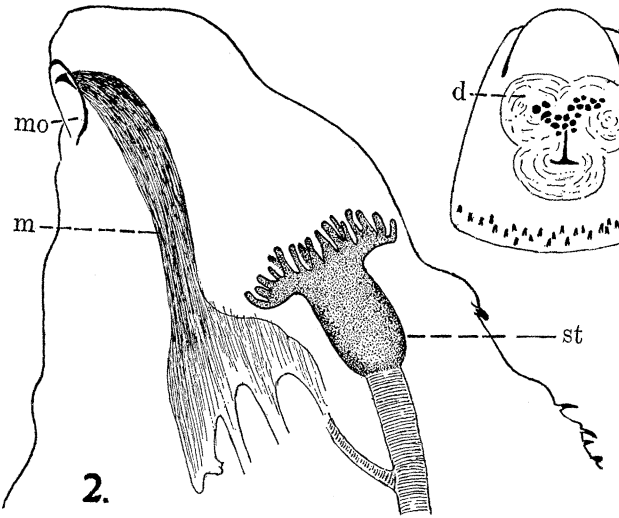

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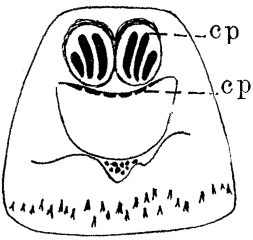

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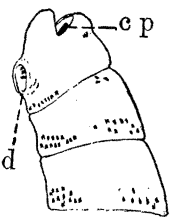

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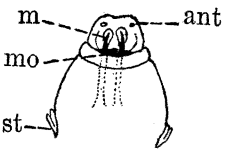

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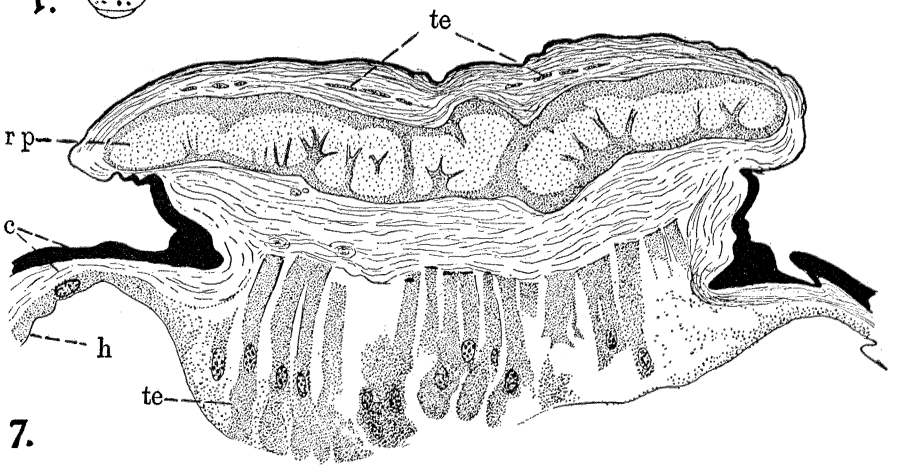

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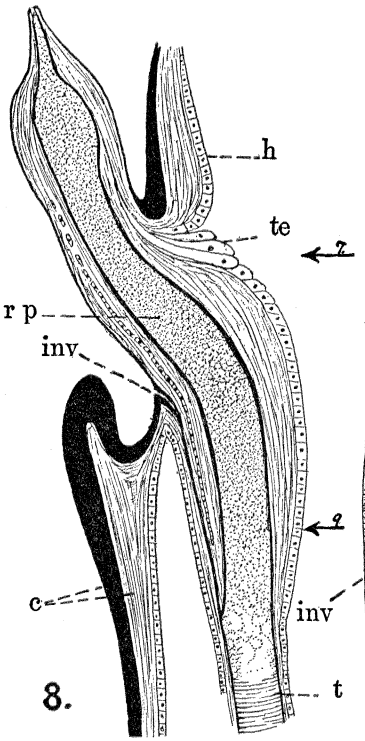
Explanation of Plate II.

Fig. 8. Reconstructed drawing of anterior stigma. *h.*, hypodermis; *te.*, tracheal epithelium; *t.*, tænidia; *c.*, cuticle; *rp.*, reticulated plug; *inv.*, invagination of cuticle. × 200.

Fig. 9. Transverse section of trachea through level indicated by arrow 9. It shows the secondary invagination with its cuticular core *inv.*, *rp.*, reticulated plug; *te.*, tracheal epithelium. × 500.

Fig. 10. Part of trachea in the transitional zone between the reticulated plug and the tænidia of the trachea. *r.*, reticulated chitin arising from the denser layer of chitin; *te.*, traceal epithelium. × 1,500.

Fig. 11. Part of wall of trachea. *t.*, tænidia; *te.*, tracheal epithelium; *tl.*, tracheal lining. × 1,500.



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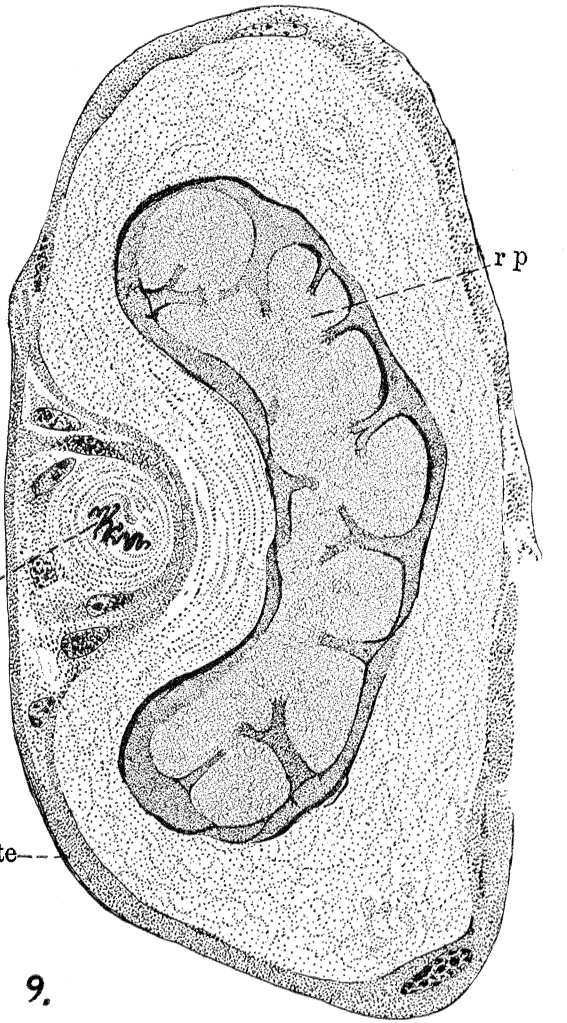

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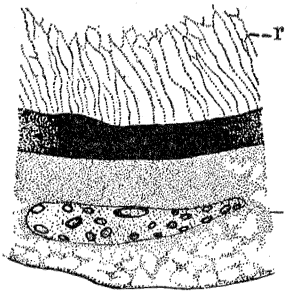

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